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STRUCTURAL CHANGES IN THE KIDNEY OF BARBUS SHARPEYI (CYPRINIDAE) YOUNGS ADAPTED TO BRACKISHWATER

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ABSTRACT

Youngs of *Barbus sharpeyi* were exposed for 14 weeks to brackishwater at a concentration of 2.1 and 2.5 ppt. The fish showed marked histopathological changes in kidney tissues, which were characterized by intracytoplasmic vacuoles of the proximal tubules with narrowing in the tubular lumen, degeneration of the distal tubules which were less influenced than proximal tubules, and contraction and melanization of glomerulus with expansion of intracapsular space. Macrophage centers were recorded in an increasing order towards the longer tested period. The kidney structure showed worthless alterations in relation to captivity in fresh water.

Keywords: Barbus sharpeyi, brackishwater, kidney, salinity, tubule, glomerulus, histopathology.

1. INTRODUCTON

The mesopotamian marshlands are a place of great environmental and cultural importance to Iraq, and were the largest in the Middle-East region. The freshwater cyprinid fish Bunni *Barbus sharpeyi* is one of the culturally important barbus species.

Due to depletion of valuable fish species in southern marshlands, some trails are carried out to furnish these marshes with fingerlings produced by artificial spawning, whereas *B. sharpeyi* is the most species compensated there.

It was investigated that the salinity approached 2.1- 2.5 ppt in several sites of east Hammar marshland during July 2006 (Hussain and Taher, 2007), which reveals brackish water characteristic. Macan and Worthington (1973) stated that the number of true brackish water species is small.

Kidney is a vital organ of body for it is important to maintain the homeostasis, maintaining volume and pH of blood and body fluids, as well as, erythropoiesis (Hickman and Trump, 1969). It is well known that freshwater teleosts keep up the hypertonicity of their blood mainly by the help of the water-excreting kidney. Several fish species can alter the rate of kidney function when subjected to different levels of salinities, for example, in trout 45% of nephrons in freshwater are filtering, whereas only 5% in seawater are filtering (Bone and Marshall, 1982). However, the kidney may undergo some structural changes which affect other functions, as ervthrocytes production, and consequently, condition of fish in general. Silva and Martinez (2007) stated that it be capable to apply the kidney histopathology as a general quality indicator of the aquatic environment. In comparison, Phromkunthong et al. (1999) observed that there were no significant differences in survival rate of yellow mystus Mystus nemurus fingerlings exposed to salinity levels from 0 to 7 ppt, but there were histological changes in kidneys of fishes raised in salinities above 5ppt, so this fish seems less sensitive to salinity than B. Sharpeyi.

It will be, therefore, interest to know the changes that kidney undergoes in *B. sharpeyi* when it is adapted to brackishwater of marsh; and this is the pioneer study on histological effects of salinity on kidney of *B. sharpeyi*.

2. MATERIALS AND METHODS

A total of 120 youngs of *Barbus sharpeyi*, measured 110-138 mm in total length, were collected from local freshwater ponds and stocked in aquaria filled with 54 litter of ancient tap-water for 10 days, and with 20 individuals in each aquarium. The fishes were fed every day with pellets. Three specimens were sacrificed to examine the natural, structural components of the kidney.

The specimens were grouped into three groups, the first and second were put in 2.1 and 2.5 ppt salinity, respectively while the third maintained as control group with a salinity of 0.7 ppt. The temperature was kept by a heater and thermostat with limits of 22-23°C. The measurements were recorded by YSI 556 MPS.

Three specimens were picked randomly from each tank weekly and dissected along 14 weeks. The kidneys were fixed in Bouin's fluid, dehydrated in alcohol, embedded in paraffin, sectioned at 6 μ m (micrometer), and stained with Mallory triple staine or Harris hematoxylin and eosin, as well as, toluidine blue (Bancroft and Stevens, 1982). Measurements of structural dimensions and density were carried out with an eyepiece micrometer and eyepiece square respectively, following Hwang and Wu (1988). Photographs were captured using the digital camera Sony DSC-T 77 (10.1 mega pixel).

3. RESULTS

Fish at all treatments survived the entire exposure period, although they were feebler at brackish water after 14 weeks of exposure compared with that at control treatment.

3.1. Control treatment

3.1.1. Gross morphology

The kidneys have the general morphological appearance, as they situated in their locality. They seemed pink in colour perior to captivity; yet they appeared as ©2006-2013 Asian Research Publishing Network (ARPN). All rights reserved.



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dark red organs with termination of research, and thiner at posterior, where they develop slight degeneration in their tubules if stocked into aquaria.

3.1.2. Histology

The various segments of nephrons and glomeruli, as well as, areas of hematopoietic tissue and blood vessels were distinguished histologically among different parts of kidney. The head kidney consisted of lymphoid, interrenal and chromaffin tissue, as the interstices of the tubules are inriched with haematopoietic tissue, which contained round to polygonal cells possessing hyper chromatic nuclei (Figure-1). Variable amounts of hematopoietic tissue and pigment cells are distributed among the vascular spaces and the tubules in the trunk kidney.

In general, it was calculated that proximal tubules I, proximal tubules II, distal tubules and glomeruli were in ratio of 2: 2: 2: 1 respectively, as there was a neck segment (Figure-1).

The glomerulus was an oval structure measured about 120 μ m in its widest diameter, as it composed of widely patent capillaries, along with some histological sections showed cluster of glomeruli accounting five ones. The walls of capillaries inside glomerulus seem bright with eosin staining. Each Bowman's capsule is connected to the archinephric duct by a short neck lined by flattened epithelium (Figure-2), as there was no distinguished space inside the capsule surrounding the glomerulus (Figures 1 and 3).

The main cells of collecting ducts are characterized by division of their cytoplasm into a dark apical half and a light basal half (Figure-3). No conspicuous distraction was observed through proximal tubules after five weeks of captivity, as they showed uniformly, pale stained cytoplasm and nuclei. However, some lysosomes were noticed inside some cells of the tubules (Figure-1).

There was a scarcity of macrophage centers, but no necrotized tissue within kidney sections at onset of captivity, as the deposition levels remained low up to 11 weeks of experiment. Twelve weeks following captivity, more macrophage centers were shown, where they attained 400 macrophage centers per mm² (Figure-4), as they seemed more prominent with toluidine blue stain. Still, the tubules and glomeruli have no obvious disorganization, where the proximal tubules were slightly influenced, while the distal tubules are still unimpaired, as well as, the blood vessels contained blood cells.

A recognizable alteration commenced 13 weeks following captivity for there were increased macrophage centers occupying hematopoietic tissue and other sites of kidney. Some glomeruli showed a degree of contraction, whereas the proximal tubules being partially destroyed; yet, the distal tubules unimpaired, for they included a few pycnotic nuclei and darker stained nuclei and cytoplasm, but the lumens are still open (Figure-5).

3.2. ppt salinity treatment

Progressive degenerative changes in kidney histology were noticed in relation to this treatment. After four weeks of exposure there was a small space inside some of Bowman's capsules around the glomerulus which showed a partial contraction in its diameter as it measured about 60 μ m. Still, some of these capsules have no such space after 11 weeks of exposure (Figure-6), and some glomeruli had disintegration of parietal layer.

A number of proximal tubules II showed disorganization with pycnotic and hyperchromatic nuclei during one week of treatment, and the intercellular spaces appeared as vacuoles within trophic cells. After four weeks the tubule cells had more uniformly pale stained cytoplasm along with pycnotic deeply stained nuclei. Certain degenerated cells were observed through these tubules during 10-11 weeks, though their lumens continue to be open or partially closed at most. Simultaneously, the proximal tubules I were less affected, but they showed darker stained cytoplasm than proximal tubules II.



Figure-1. Cross section of the kidney from control treatment. g, glomerulus; P1, proximal tubule I; P2, proximal tubule II; d, distal tubule; h, hematopoietic tissue; n, neck. (H and E)*. × 400.



Figure-2. Cross section of the kidney showing the bowman's capsule (g) connected by a neck lined by flattened epithelium (f). (Mallory).** × 600.

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Figure-3. Cross section of the kidney showing the glomerulus (g) without a space around it. c, collecting duct; p, proximal tubule; d, distal tubule; h, hematopoietic tissue. (H and E). × 200.



Figure-4. Cross section of the kidney after 12 weeks in control treatment showing some macrophage centers (m). The proximal tubules (p) are slightly influenced, while the distal tubules (d) are still unimpaired. Toluidine bule stain. $\times 100. * H$ and E = hematoxylin and eosin stain; ** mallory triple stain.



Figure-5. Cross section of the kidney after 13 weeks in control treatment showing some macrophage centers among hematopoietic tissue (h), contracted glomerulus (g), destroyed proximal tubules (p) and partial destroyed distal tubules (d). (Mallory). × 200.



Figure-6. Cross section of the kidney after 11 weeks of 2.1 ppt treatment showing ordinary glomerulus (g) disorganized proximal tubule (p), lymphocytes crossing distal tubule (d1), distal tubule with darked stained and pycnotic nuclei (d2) and some macrophages (c) and mucous cells (m). (H and E). × 300.

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Figure-7. Cross section of the kidney of 2.1 ppt treatment after 12 weeks showing two disintegrated glomeruli (g), hemorrhage (h). p, disorganized proximal tubule; d, distal tubule; n, necrosis. (Mallory). × 400.



Figure-8. Cross section of the kidney of 2.1 ppt treatment after 13 weeks showing the degenerated glomerulus (g), destroyed proximal tubules (p), necrotic hematopoietic tissue (h), neck segment (n) and increased macrophage centers (m). (H and E). × 300.

The distal tubules, up to 11 weeks of exposure, were mostly retained their general architecture, as they characterized by open lumen with darker stained cytoplasm and nuclei in comparison with disorganized proximal tubules. Some of distal tubules contained lymphocytes crossing their cells, as there were more macrophages and mucous cells could be noticed across the examined sections.

At twelfth week of exposure a number of proximal tubules underwent a complete destruction, so the two types of them couldn't be distinguished, whereas another disorganized in different levels, as they contained more of degenerated or vacuolated cells, even the nuclei, in general, exhibited pycnotic shape and darker stain as reflected by the three stains employed. Also, a few separated spots of hemorrhage were formed, along with some blotches of necrosis formed at that time (Figure-7).

The melano-macrophages appeared as dark brown or black centers which increased progressively in number and size with continuation of exposure period, whether in control treatment (Figure-4), or in salinity treatment. The noticeable appearance was observed after 11 weeks of exposure for there was about 400 macrophage center per mm² (Figure-8). Majority of such centers scattered through the necrotic hematopoietic tissue along with some phagocytes and mucous cells, as well as, through archinephric duct which full with blood cells.

The actual destruction of kidney occurred after 13 weeks of treatment where most proximal tubules showed disorganization or exhibited closed lumen. Simultaneously, some of distal tubules were obviously degenerated, whereas the remainder have increased pycnotic nuclei with constricted or closed lumen; still, few of them retained the characterized general appearance. The glomeruli, in turn, started obvious degeneration, as they showed impaired vessels containing residue of blood cells. The hematopoietic tissue comprised more pycnotic nuclei with vast necrosis, besides; there were increased macrophage centers (Figure-8).

3.3. ppt salinity treatment

It was noticeable that distal tubules were affected during four weeks of treatment in this salinity compared with 11 weeks in previous treatment. They comprised some lysosomes inside cells, along with a number of pycnotic dark stained nuclei which may amounted 1/4 of total nuclei; however, they were retaining open lumen (Figure-9). Cytoplasm of distal tubules epithelium seemed darker and more uniform than that of proximal tubules. On the other hand, several proximal tubules appeared with tight or closed lumen during four weeks of treatment, besides, a certain cells showed vacuolated cytoplasm and gaint nucleus. The initial stage in the degeneration employment marked by the formation of eosinophilic granules inside the cells, in addition to lysosomes. Some macrophage centers and melanized glomeruli are diffused across the tubules.

Glomeruli, in turn, being influenced during five weeks of treatment since some of them showed a constriction inside Bowman's capsule and has an average $55 \mu m$ in diameter, as some of them are being elongated. Anyway, the glomeruli retained the favoured form and contained blood cells within vessels up to 11 weeks of exposure (Figure-10), and certain ones appeared melanized.

Six or seven weeks following exposure there was noticeable necrosis among hematopoietic tissue with pycnotic nuclei at head kidney. In trunk kidney the effective damage appeared as regions of necrosis during 11 weeks; however, the peritubular capillaries are mostly VOL. 8, NO. 4, APRIL 2013

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filled by blood cells, but the neck segments showed pale staining (Figure-11).

Arriving to 13 weeks, the treatment resulted in disintegrated glomeruli which seem to be feeble structures in filtration rate. Most of proximal tubules were destroyed, but the distal ones are less affected. Vast areas of hematopoietic tissue were occupied by melano-macrophages with phagocytes and mucous cells (Figure-12).

There is evidence that *B. sharpeyi* has the ability to develop new nephrons following renal injury, although it seems that it has less ability after 13 weeks of exposure.

It seems that salinity effects are fare more pronounced in acute conditions for some components of kidney (such as distal tubules), whereas more pronounced in chronic exposure for others (such as glomeruli). So, the fishes in both concentrations of salinity appeared feeble swimmers at 14 th weeks, whereas they were more active at control treatment.

4. DISCUSSIONS

The three tubules (proximal I, proximal II and distal) are present in approximate proportion in the kidney of the freshwater fish *B. sharpeyi*, as it is endemic species in Mesopotamica (khalaf, 1961; Beckman, 1962). Kato *et al.* (2005) suggested that the presence of distal segment in kidney indicates a freshwater adaptation, where they act as urine-diluting segment, thus the presence of such segment is one of the most important factors that allow *Takifugu osbcurus* to be highly adaptable to a wide range of salinities.



Figure-9. Cross section of the kidney of 2.5 ppt treatment showing vacuolated cytoplasm inside proximal tubule (v), lymphocytes crossing distal tubule (1), macrophage centers (m) and melanized glomerulus (g). (Mallory). × 400.



Figure-10. Cross section of the kidney showing expansion of intercapsular space (s), around the elongated glomerulus (g). (H and E). \times 1000.



Figure-11. Cross section of the trunk kidney of 2.5ppt treatment showing necrotic regions (c), destroyed proximal tubules (p) and neck segment with pale stain (n). (H and E). × 1000.



Figure-12. Cross section of the head kidney of 2.5 ppt treatment after 13 weeks of exposure showing the destroyed tissue. (Mallory). × 1000.

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Sakai (1985) noticed that the main cells of distal tubules contain abundant mitochondria, but have no brush border. Glomerulus diameter in *B. Sharpeyi* youngs (120 μ m) also indicates a freshwater adaptation (Lagler *et al.*, 1970), the freshwater or brackish water adapted juveniles of tarpon *Megalops atlantica* possessed a glumeruli attaining at most a diameter of 40 μ m, although it was recognized that the results are not statistically valid (Rasquin, 1969). On the other hand, Perez *et al.* (2003) deemed the aglomerularism in *Harpagifer bispinis*, a modification that contributing to survival in a hyposmotic medium.

The mode of connection observed in Bowman's capsule may suggest primitive situation which remarks this cyprinid fish in relation to kidney structure, as it is pronounced in primitive taxon Hagfish (Hickman and Trump, 1969). However, the present species is characterized by the neck segment which is absent from some cyprinds like *C. auratus* (Sakai, 1985).

Macrophage centers are a result of captivity stress or due to salinity treatment, they can be increased due to chemicals exposure as observed in Cyprinus carpio after acut exposure to deltamethrin (Cengiz, 2006), as well as, due to starvation as observed in olive flounder Paralichthys olivaceus (Hur et al., 2006) or in mugil fish Liza abu (Jasim, 2006). It could be assumed that the head kidney was more influenced than trunk kidney as long as the necrotized tissue and macrophage centers were mostly noticed at hematopoietic tissue. The lysosomes inside cytoplasm of the tubules, in turn, an expression of physiological activity, where Hickman and Trump (1969) interpreted it as acid phosphate activity in southern flounder. Likewise, the presence of eosinophilic granules inside tubules cells points out to degeneration process for them, and afterwards, the degeneration of kidney.

The uniformly stained cytoplasm in the tubules of control specimens is the status of freshwater adapted fish. Virabhadrachari (1961) observed a similar situation in *E. maculates* adapted to freshwater, which showed asymmetric cytoplasm when adapted to 30% seawater. Also, the division of cytoplasm in collecting duct into dark and light halves was observed in *C. auratus* (Sakai, 1985).

The reduction of glomerulus area when exposed to brackishwater seems to be related to an abated filtering role, as it was noticed in some freshwater species such as O. niloticus and O. mossambicus when transferred to higher salinity (Hwang and Wu, 1988; Cataldi et al., 1991). Glomerulus in B. sharpeyi being influenced during 4-5 weeks in both treatments of salinity, although it showed more contraction in the course of 2.5 ppt salinity. In contrast, the glass eels A. anguilla showed a better growth performance and nutritional condition when fed hake roe, independently of the salinity ranged from 0-15 ppt (Rodriguez et al., 2005). It is logical to assume that the total filtering surface is decreased with reduction of the glomerular size, besides, the intercapsular space showed an enlargement with glomerular atrophy. Such space was observed in P. lineatus under Trichlorfon pollution exposure (da Veiga et al., 2002) and C. mrigala when exposed to sub-lethal concentrations of fenvalerates (Velmurugan *et al.*, 2007). Likewise, the several smashing glomeruli after 13 weeks of treatment indicate a dysfunction of them. Elger and Hentschel (1981) observed that more than 90% of the glomeruli disappeared from tissue in stenohaline freshwater teleost *C. auratus gibelio* adapted to 15ppt salinity.

The change in kidney morphology could be supposed as osmoregulatory strategies that to withstand facing osmotic alteration. The intercellular spaces formed at the epithelium of proximal tubules with decrease of lumen diameter seem a response to salinity stress. Perez et al. (2003) observed similar situation in subantractic fish H. bispinis acclimated at 2ppt salinity, and justified that as a modification which facilitate urine secretion, thus contributing to the survival in hyposmotic medium, Velmurugan et al. (2003) observed a decrease in tubule lumen as a response to fenvalerate pollution. Virabhadrachari (1961) noticed a decrease of lumen in E. maculates adapted to 30% seawater, likewise, Brown et al. (1980) noticed a reduced filtering tubules of S. giardneri in seawater compared with that in freshwater. It was suggested that internal cells hypertrophy in Astyanax altiparanae_indicates a chronically exposure to stressors in the environment (Silva and Martinez, 2007). In trout, it was noticed that 45% of nephrons in freshwater are filtering, whereas only 5% in seawater are filtering (Bone and Marshall, 1982). However, the proximal tubules were more affected than distal ones at all treatments as they being destructed perior to distal tubules or glomeruli.

These results suggest that the degree of melanomacrophage centers deposition in kidney could be utilized as alternative indicator to identify salinity stress on *B. sharpeyi*. da Veiga *et al.* (2002) concluded that "morphologic kidney changes may induce defence system changes harming the animals homeostasis and healthy".

It is concluded that *B. sharpeyi* is less tolerant than some fishes inhabiting freshwater such as tilapia *O. niloticus* and *O. mossambicus* which adapted to 35ppt, although they had reduction in glomerulus size and change in tubules morphology (Cataldi *et al.*, 1991). So, the culturing of Bunni in brackish water can cause significant damage to the general health of fish.

The ability of Bunni to develop new nephrons following renal injury has shown in some species as Atlantic tomcod and brown bullhead (Cormier *et al.*, 1995).

5. CONCLUSIONS

Youngs of Bunni *B. sharpeyi* seem to be sensitive to reduced levels of salinity as realized by histological changes in kidney. The distal tubules being more resistive than the proximal ones, and the head kidney is more influenced than the trunk kidney, as the kidney in general, indicates a freshwater properties. This must be considered in projects of culture concerning this fish.

The reduction of glomerulus size or the formation of intercellular spaces at proximal tubules epithelium being in accordance with salinity increase, as it was seen



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to be associated with pollution exposure at several fishes studied.

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REFERENCES

Beckman W. C. 1962. Freshwater fishes of Syria and their general biology and management. FAO. Fisheries biology branch, Technical paper No. 8.

Bone Q and N.B. Marshall. 1982. Biology of Fishes. Blackie, Glasgow, Scotland.

Brown A. J., J.A. Oliver, I.W. Henderson and B.A. Jackson. 1980. Angiotensin and single nephron glomerular function in the trout *Salmo gairdneri*. A. J. Physiol. 239: R 509-R 514.

Cataldi E., L. Garibaldi, D. Crosetti, C. Leoni and S. Cataudlla. 1991. Variations in renal morphology during adaptation to salinities in tilapias. Environ. Bio. Fish. 31: 101-106.

Cengiz E. I. 2006. Gill and kidney histopathology in the freshwater fish *Cyprinus carpio* after acute exposure to deltamethrin. Environ. Tox. Pharm. 22: 200-204.

Cormier S. M., T.W. Neiheisel, P. Wernsing, R.N. Racine and R. Reimschuessel. 1995. New nephron development in fish from polluted waters: a possible biomarker. Ecotoxicology. 4: 157-168.

da Veiga M.L., E.de L. Rodrigues, F.J. Pacheco and M.J.T. Ranzani-Paiva. 2002. Histopathologic changes in the kidney tissue of *Prochilodus lineatus* Valenciennes, 1836 (Characiformes, Prochilodontidae) induced by sublethal concentration of trichlorfon exposure. Braz. Arch. Boil. Technol. 45: 1-7.

Elger M. and H. Hentschel. 1981. The glomerulus of a stenohaline freshwater teleost, *Carassius auratus gibelio*, adapted to Saline water. Cell tiss. Res. 220: 73-85.

Hickman C. P. Jr. and B.F. Trump. 1969. The Kidney. In: Fish Physiology. Hoar, W.S. and D. J. Randall (Eds.). Academic Press, New York, USA. 1: 91-239.

Hur J. W., S.R. Woo, J.H. JO and I-S Park. 2006. Effects of starvation on kidney melano-macrophage centre in olive flounder, *Paralichthys olivaceus* (Temminck and Schlegel). Aqua. Res. 37: 821-825.

Hussain N. A. and M.A. Taher. 2007. Effect of daily variations, diurnal fluctuations and tidal stage on water parameters of East Hammar marshland, Southern IRAQ. Marsh Bulletin. 2: 32-42.

Hwang P. P. and S.M. WU. 1988. Salinity effects on cytometrical parameters of the kidney in the euryhaline teleost *Oreochromis mossambicus* Peters. J. Fish Biol. 33: 89-95.

Jasim B. M. 2006. Effect of starvation stress on organ structure in mugil fish *Liza abu*. Basrah J. Sci (B). 24: 86-102.

Kato A., H. Doi, T. Nakada, H. Sakai and S. Hirose. 2005. *Takifugu obscurus* is a euryhaline fugu species very close to *Takifugu rubripes* and suitable for studying osmoregulation. BMC Physiology. 5: 18-28.

Khalaf K. T. 1961. The Marine and Fresh Water Fishes of Iraq. Ar-Rabitta Press, Baghdad, Iraq.

Lagler K. F., J.E. Bardach, R.R. Miller and D.R.M. Passino. 1970. Ichthyology. John Willey and Sons, New York, USA. p. 506.

Macan T. T. and E.B. Worthington. 1973. Life in Lakes and Rivers. Collins, the Fontana New Naturalist, London, U.K.

Perez A. F., J. Calvo, M. Tresguerres and C. Luquet. 2003. Aglomerularism in *Harpagifer bispinis*: a subantarctic notothenioid fish living at reduced salinity. Polar biology. 26: 800-805.

Phromkunthong W., K. Supamattaya, K.Saelee and P.Torrarit. 1999. Effect of salinity levels on growth performance, physiological and histological changes in yellow mystus, *Mystus nemurus*. Songklanakarin J. Sci. Tech. 21(1): 53-64.

Rasquin P. 1969. Undescribed glands in the kidney of the juvenile tarpon *Megalops atlantica*. Copeia. No. 1: 83-91.

Rodriguez A., E. Gisbert, G. Rodriguez and F. Castello-Orvay. 2005. Histopathological observations in European glass eels (*Anguilla anguilla*) reared under different diets and salinities. Aquaculture. 244(1-4): 203-214.

Sakai T. 1985. The structure of the kidney from the freshwater teleost *Carassius auratus*. Anatomy and Embryology. 171: 31-39.

Silva A. G. and C.B.R. Martinez. 2007. Morphological changes in the kidney of a fish living in an urban stream. Env. Tox. Pharm. 23: 185-192.

Velmurugan B., M. Selvanayagam, E.I. Cengiz and E. Unlu. 2007. The effects of fenvalerate on different tissues of freshwater fish *Cirrhinus mrigala*. J. Env. Sci. Heal. (B). 42: 157-163.

Virabhadrachari V. 1961. Structural changes in the gills, intestine, and kidney of *Etroplus maculatus* (Teleostei) adapted to different salinities. Quar. J. mic. Sci. 102: 361-369.